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EXAMINER	
STEADMAN, DAVID J	

ART UNIT	PAPER NUMBER
1652	

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Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/044,303	MANSTEIN ET AL.	
	Examiner	Art Unit	
	David J Steadman	1652	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *27 August 2003*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1,3,4,6,8-13,28 and 30-32* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1,3,4,6,8-13,28 and 30-32* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. §§ 119 and 120

12)☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☒ All b)☐ Some * c)☐ None of:

1.☒ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

13)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application) since a specific reference was included in the first sentence of the specification or in an Application Data Sheet. 37 CFR 1.78.

a) ☐ The translation of the foreign language provisional application has been received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121 since a specific reference was included in the first sentence of the specification or in an Application Data Sheet. 37 CFR 1.78.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO-1449) Paper No(s) _____.
4) ☐ Interview Summary (PTO-413) Paper No(s). _____.
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: .

DETAILED ACTION

Status of the Application

[1] Claims 1, 3-4, 6, 8-13, 28, and 30-32 are pending in the application.

[2] Applicants' amendment to the claims filed August 27, 2003 is acknowledged. This amendment replaces all previous versions and listings of the claims in the instant application.

[3] Receipt of a corrected Declaration filed August 27, 2003 is acknowledged.

[4] Applicants' arguments filed August 27, 2003 have been fully considered and are deemed to be persuasive to overcome some of the rejections and/or objections previously applied. Rejections and/or objections not reiterated from previous office actions are hereby withdrawn.

[5] The text of those sections of Title 35 U.S. Code not included in the instant action can be found in a prior Office action.

Claim Objections

[6] In view of applicants' amendment to claims 6 and 11, the objections to the claims as set forth in items 5-6 of the Office action mailed February 25, 2003, are withdrawn.

Claim Rejections - 35 USC § 112, Second Paragraph

[7] In view of applicants' amendment to claims 10 and 12, the rejection of the claims under 35 U.S.C. § 112, second paragraph, as set forth in items 7a-b of the Office action mailed February 25, 2003, are withdrawn.

Claim Rejections - 35 USC § 112, First Paragraph

[8] The written description rejection of claims 1, 3-4, 6, 8-10, 13, 28, and 30-32 under 35 U.S.C. 112, first paragraph, is maintained for the reasons of record as set forth in item 8 of the Office action mailed February 25, 2003 and the reasons stated below. Applicants' arguments addressing the instant rejection begin at page 6 of the response filed August 27, 2003. It should be noted that applicants address the written description and scope of enablement rejections collectively and not individually. To the extent

applicants' arguments address the instant rejection, applicant's arguments are traversed below. At the top of page 6, applicants argue claim 1 has been amended and that in view of the amendment the rejection should be withdrawn. Applicants argue that a representative species of the claimed genus is described in the specification, along with other examples that had undergone various degrees of testing at the time of the invention. Applicants argue this represents a representative number of the claimed genus of recombinant proteins to show applicants were in possession of the claimed invention. Applicants' arguments are not found persuasive.

In this case, only a single representative species of the claimed genus of recombinant proteins has been described in the specification, i.e., a recombinant protein comprising SEQ ID NO:1 and a target protein of interest fused by a linker. It should be noted that all of applicants' asserted representative species include SEQ ID NO:1 as part (a) of claim 1. The specification makes clear that the protein analog of part (a) of claim 1 can have a change in biological activity or structure of the native sequence (page 8, paragraph [0027]). Therefore, the first protein of claim 1 is not limited to a protein having the structure or function of myosin, but broadly encompasses any protein structure or function. Contrary to applicants' assertions, the single disclosed representative species fails to describe the entire genus of claimed recombinant proteins. Part (a) of claim 1, *i.e.*, "a first protein that is a motor domain of myosin, or an analog, fragment or derivative thereof", encompasses widely variant species, including any protein having any structure and any function. While MPEP § 2163 acknowledges that in certain situations "one species adequately supports a genus", it is also acknowledges that "[f]or inventions in an unpredictable art, adequate written description of a genus which embraces widely variant species cannot be achieved by disclosing only one species within the genus". As stated in a previous Office action, the claimed genus encompasses a vast number of species with widely variant structures and functions. As such, the disclosure of the single representative species of a recombinant protein comprising SEQ ID NO:1 and a target protein of interest fused by a linker is insufficient to be representative of the attributes and features of *all* species encompassed by the claimed genus of recombinant proteins. Given the lack of description of a representative number of recombinant proteins as encompassed by the claims, the specification fails

to sufficiently describe the claimed invention in such full, clear, concise, and exact terms that a skilled artisan would recognize that applicant was in possession of the claimed invention.

[9] The scope of enablement rejection of claims 1, 3-4, 6, 8-10, 13, 28, and 30-32 under 35 U.S.C. 112, first paragraph, is maintained for the reasons of record as set forth in item 9 of the Office action mailed February 25, 2003 and the reasons stated below. Applicants' arguments addressing the instant rejection begin at page 6 of the response filed August 27, 2003. It should be noted that applicants address the written description and scope of enablement rejections collectively and not individually. To the extent applicants' arguments address the instant rejection, applicant's arguments are traversed below. At the top of page 6, applicants argue claim 1 has been amended and that in view of the amendment the rejection should be withdrawn. Applicants argue that a working example of the claimed invention is described in the specification, along with other examples that had undergone various degrees of testing at the time of the invention. Applicants argue the success rate in expressing the recombinant protein of the invention was nearly 70%. Applicants argue 80% of the proteins were purified and two of four were crystallized and their structures determined. Applicants argue the specification contains ample guidance for a skilled artisan to make and use the entire scope of recombinant proteins without undue experimentation. Applicants' arguments are not found persuasive.

The examiner maintains his position that undue experimentation would be required to make and use the entire scope of claimed recombinant proteins and protein crystals for those reasons as set forth in detail at pages 6-8 of the Office action mailed February 25, 2003. While applicants have amended part (a) of claim 1, the amendment does not limit the scope of the claim to the enablement provided by the specification. Claim 1 part (a) broadly encompasses any protein having any structure and any function, including proteins that have yet to be isolated and claim 28 broadly encompasses any protein crystals of the recombinant protein of claim 1. The specification provides only a single working example of the protein of part (a) of claim 1 – SEQ ID NO:1. This single working example fails to provide the guidance necessary for making and using *all* proteins broadly encompassed by the claims. Even those fusion proteins having SEQ ID NO:1 or fragments thereof have an inherently high degree of unpredictability in their expression as the specification discloses that nearly one-third of fusion proteins having SEQ ID

NO:1 as a fusion partner were not expressed by a host cell (see page 29, Table 1 of the specification). Regarding making crystals of the broad scope of claimed recombinant proteins, it is noted that Branden et al. ("Introduction to Protein Structure, Second Edition", Garland Publishing Inc., New York, 1999) teach that "[p]rotein crystals are difficult to grow" (page 374) and provide reasons to support the examiner's argument that undue experimentation would be required to make the entire scope of claimed protein crystals. Thus, in view of the overly broad scope of the claims, the lack of guidance and working examples provided in the specification, and the high degree of unpredictability as evidenced by the specification or prior art, undue experimentation would clearly be necessary for a skilled artisan to make and use the entire scope of the claimed invention.

Claim Rejections - 35 USC § 102

[10] The rejection of claims 1, 6, and 13 under 35 U.S.C. § 102(e) as being anticipated by Finer et al. (US Patent 6,410,254) is maintained for the reasons of record as set forth in item 10 of the Office action mailed February 25, 2003 and the reasons stated below. Applicants' arguments addressing the instant rejection begin at page 7 of the response filed August 27, 2003. Beginning at the top of page 8, applicants argue Finer et al. do not teach a fusion protein having a catalytic domain of myosin, linked by an oligopeptide linker to a target protein of interest, the purification and crystallization of which is desired. Applicants' argument is not found persuasive.

Applicants' appear to assert that the reference of Finer et al. is not enabling to anticipate the claimed invention as Finer et al., while teaching all limitations of the claimed invention (for detailed teachings of Finer et al., see page 9, item 10 of the Office action mailed February 25, 2003), allegedly do not teach a specific myosin or a specific embodiment of a fusion protein. It should be noted that the claims are not so limited to a fusion protein having a catalytic domain of myosin as argued by applicant. In this case, claim 1 recites, "a first protein that is a motor domain of myosin, or an analog, fragment or derivative thereof". It should also be noted that the limitation of "the purification and crystallization of which is desired" for the target protein in no way limits the target protein of part (b) of claim 1. A recitation of the intended use of the claimed invention must result in a structural difference between the claimed

invention and the prior art in order to patentably distinguish the claimed invention from the prior art. If the prior art structure is capable of performing the intended use, then it meets the claim. Finer et al. clearly teach a fusion comprising the catalytic domain of myosin as stated in the previous Office action (see columns 17-18 of Finer et al.). At the time of the invention, methods of making recombinant fusion proteins, including those comprising myosin proteins known in the art, were known in the art as acknowledged by Finer et al. (column 18, lines 23-24) and applicants have presented no evidence that would suggest otherwise. See MPEP §§ 2121 and 716.07. Thus, Finer et al. anticipate the claimed invention.

[11] In view of applicants' amendment to claim 1 to recite the limitation of "a linker between (a) and (b)", the rejection of original claims 1, 4-7, 9-10, and 13 under 35 U.S.C. § 102(b) as being anticipated by Kurzawa et al. (*Biochemistry* 36:317-323, 1997) as set forth in item 11 of the Office action mailed February 25, 2003, is withdrawn. Kurzawa et a. do not teach the presence of a linker joining the myosin and alpha-actinin components of their fusion protein.

[12] In view of applicants' amendment to claim 1 to recite the limitation of "a linker between (a) and (b)", the rejection of original claims 1, 4-7, 9-10, and 13 under 35 U.S.C. § 102(b) as being anticipated by Manstein et al. (*J Mus Res Cell Mot* 16:325-332; IDS reference CF) as set forth in item 12 of the Office action mailed February 25, 2003, is withdrawn. Manstein et al. (*J Mus Res Cell Mot* 16:325-332) do not teach the presence of a linker joining the myosin and alpha-actinin components of their fusion protein.

[13] Claims 1, 3-4, 6, 8-9, and 30-31 are rejected under 35 U.S.C. 102(b) as being anticipated by Wolber et al. (*Biotechnology* 10:900-904). This rejection is necessitated by amendment. The claims are drawn to a recombinant protein comprising a first protein that is a motor domain of myosin or an analog, fragment or derivative thereof, a target protein of interest, and a linker between the two protein components. Claims 30-31 limit the linker to comprising (in relevant part) a factor Xa cleavage site. Wolber et al. teach an expressed fusion protein comprising a fragment from the rod portion of myosin heavy chain, light meromyosin (LMM) from rabbit skeletal muscle (page 900, right column, bottom) linked to a p21, NF1, HIV-1 Tat or HIV-1 protease protein via a linker comprising a factor Xa cleavage site (page 901, right column to page 902, left column). Wolber et al. teach the amino acid sequence of the factor Xa

cleavage site is Ile-Glu-Gly-Arg (page 900, bottom). This anticipates claims 1, 3-4, 6, 8-9, and 30-31 as written.

[14] Claim 1, 3-4, 6, 8-9, 28, and 30-31 are rejected under 35 U.S.C. 102(b) as being anticipated by Kuge et al. (*Prot Sci* 6:1783-1786). This rejection is necessitated by amendment. The claims are drawn to a recombinant protein comprising a first protein that is a motor domain of myosin or an analog, fragment or derivative thereof, a target protein of interest, and a linker between the two protein components. Claims 30-31 limit the linker to comprising (in relevant part) a thrombin cleavage site. Kuge et al. teach an expressed fusion protein comprising GST fused to the DNA binding domain of DREF via a linker comprising a thrombin cleavage site (page 1784, right column, bottom). Kuge et al. teach the amino acid sequence of the thrombin cleavage site is Asp-Leu-Val-Pro-Arg-Gly-Ser (page 1784, left column, bottom). Kuge et al. teach crystallization of their fusion protein (page 1784, right column). This anticipates claims 1, 3-4, 6, 8-9, 28, and 30-31 as written.

Claim Rejections - 35 USC § 103

[15] The rejection of claims 3, 4, and 8 under 35 U.S.C. § 103(a) as being unpatentable over Finer et al. in view of Bulow et al. (*Trends Biotech* 9:226-231) and Argos (*J Mol Biol* 211:943-958) is maintained for the reasons of record as set forth in item 14 of the Office action mailed February 25, 2003 and the reasons stated below. It is noted that the examiner failed to cite Bulow et al. and Argos as secondary references at the beginning of the rejection. However, as the teachings of Bulow et al. and Argos are expressly disclosed in the body of the rejection, it is clear that the rejection is made in view of Bulow et al. and Argos.

Applicants argue Finer et al. do not teach a fusion protein having a catalytic domain of myosin, linked by an oligopeptide linker to a target protein of interest. Applicants argue one of ordinary skill in the art would not have combined the cited references because: the linker has to be chosen to avoid transfer of conformational energy during crystallization and a particularly preferred linker is one that contains a proteolytic cleavage site. Applicants' argument is not found persuasive.

It is noted that the claims are not so limited to a linker comprising a proteolytic cleavage site as argued by applicants. Furthermore, the limitation of "the purification and crystallization of which is desired" for the target protein in no way limits the target protein of part (b) of claim 1. A recitation of the intended use of the claimed invention must result in a structural difference between the claimed invention and the prior art in order to patentably distinguish the claimed invention from the prior art. If the prior art structure is capable of performing the intended use, then it meets the claim. As stated above, Finer et al. anticipate the recombinant protein of claims 1, 6, and 13 and Finer et al. clearly teach the use of a linker for joining the myosin component and second polypeptide component of their fusion protein (see column 17, lines 62-64) and Bulow et al. and Argos clearly teach the advantages and desired composition of such a linker. As such, one of ordinary skill in the art would have combined the teachings of the cited references to make the claimed invention.

[16] Claims 1, 3-4, 6, 8-10, and 13 are rejected under 35 U.S.C. 103(a) as being unpatentable over Kurzawa et al. in view of Bulow et al. and Argos. This rejection is necessitated by amendment. The claims are drawn to a recombinant protein comprising a first protein that is a motor domain of mysosin or an analog, fragment, or derivative thereof; a target protein of interest; and a linker between the first protein and target protein.

Kurzawa et al. teach myosin head fragments M754 (amino acids 1-754 of *Dictyostelium discoideum* myosin II) or M765 (amino acids 1-765 of *D. discoideum* myosin II) fused to one or two alpha-actinin repeats (page 317, right column, bottom and page 318, left column, bottom). Kurzawa et al. teach the fusion proteins have a histidine octamer at the C-terminus (page 318, right column, top). Kurzawa et al. do not specifically teach a linker between their fused protein domains.

At the time of the invention, the use of oligopeptide linkers for joining two protein components in a fusion protein to allow conformational flexibility of the protein components was well known to one of ordinary skill in the art. For example, Bulow et al. teach the presence of peptide linkers between protein components of a fusion enzyme. Bulow et al. advise using linkers that are short (between two and ten amino acids) as being optimal as longer linkers are often prone to proteolytic cleavage and reduce recombinant protein yields (page 230, left column). Also, Argos teaches that oligopeptide linkers are used

in the fusion of proteins and should generally not interfere with their function (page 943, left column). Argos teaches the advantages of using oligopeptide linkers comprising Ser, Gly, and Thr as these residues can impart flexibility and maintain stability and conformation in solution (page 956, left column).

Therefore, it would have been obvious to one of ordinary skill in the art to link M761 and the alpha-actinin repeats of Kurzawa et al. using a linker comprising at least two amino acids and glycine. One would have been motivated for the fusion protein of Kurzawa et al. with a linker peptide comprising at least 2 amino acids and glycine because of the teachings of Bulow et al. and Argos. One would have a reasonable expectation of success for the fusion protein of Kurzawa et al. with a linker peptide comprising at least 2 amino acids and glycine because of the results of Kurzawa et al., Bulow et al., and Argos. Therefore, claims 1, 3-4, 6, 8-10, and 13, drawn to recombinant proteins as described above would have been obvious to one of ordinary skill in the art.

Applicants argue (page 11) that none of the cited references provides motivation for a linker to prevent transfer of conformational energy (thereby facilitating crystallization) and therefore, it would not have been obvious to combine the cited references. Applicants' argument is not found persuasive.

The linker recited in the claims is not limited to preventing transfer of conformational energy. A recitation of the intended use of the claimed invention must result in a structural difference between the claimed invention and the prior art in order to patentably distinguish the claimed invention from the prior art. If the prior art structure is capable of performing the intended use, then it meets the claim. While Kurzawa et al. do not specifically teach a linker between the myosin head fragment and the alpha-actinin repeats, clearly such linkers were well known in the art as evidenced by Bulow et al. and Argos. As such, one of ordinary skill in the art would have combined the teachings of the cited references to make the claimed invention.

[17] Claim 11 is rejected under 35 U.S.C. 103(a) as being unpatentable over Kurzawa et al. in view of Bulow et al. and Argos as applied to claims 1, 3-4, 6, 8-10, and 13 above, and further in view of Van Dijk et al. (*Eur J Biochem* 260:672-683), Ponomarev et al. (*Biochemistry* 39:4527-4532), Furch et al. (*Biochemistry* 37:6317-6326), and Van Dijk et al.(*Biochemistry* 38:15078-15085). This rejection is necessitated by amendment. Claim 11 limits part (a) of claim 1 to a protein comprising SEQ ID NO:1.

Kurzawa et al., Bulow et al., and Argos disclose the teachings as stated above. In addition, Kurzawa et al. teach that "the recombinant nature and the fad that they can be produced and purified in large amounts make M761, M761-1R, and M761-2R ideal constructs for systematic studies of the structure, kinetics, and function of the myosin motor" (page 323). The references of Kurzawa et al., Bulow et al., and Argos do not teach SEQ ID NO:1 (amino acids 1-765 of *D. discoideum* myosin II) as the myosin head fragment of their fusion protein.

At the time of the invention, it was well known in the art that M765, a myosin head fragment of amino acids 1-765 of *D. discoideum* myosin II, was useful in the characterization of the structure, kinetics, and function of the myosin motor. Van Dijk et al. (*Eur J Biochem* 260:672-683) teach M765 is the motor domain of *D. discoideum* myosin II and has been shown to retain normal ATP-hydrolysis and actin binding activities (page 672, right column, bottom). Numerous references describe the use of M765 in the structural, kinetic, and functional characterization of the myosin motor - see for example, Van Dijk et al. (*Eur J Biochem* 260:672-683), Ponomarev et al., Furch et al., and Van Dijk et al. (*Biochemistry* 38:15078-15085). Thus, it is clear from the prior art that M765 was useful in the characterization of the structure, kinetics, and function of the myosin motor.

Therefore, it would have been obvious to one of ordinary skill in the art to use M765 (amino acids 1-765 of *D. discoideum* myosin II as the myosin head fragment in the fusion protein of Kurzawa et al. One would have been motivated for the fusion protein of Kurzawa et al. with M765 instead of M761 because M765 was well characterized and established for use in the characterization of the structure, kinetics, and function of the myosin motor. One would have a reasonable expectation of success for the fusion protein of Kurzawa et al. with M761 replaced with M765 because of the results of Kurzawa et al., Van Dijk et al. (*Eur J Biochem* 260:672-683), Ponomarev et al., Furch et al., and Van Dijk et al. (*Biochemistry* 38:15078-15085). Therefore, claim 11, drawn to the recombinant protein as described above would have been obvious to one of ordinary skill in the art.

Applicants argue (page 11) that none of the cited references teaches a recombinant protein comprising a myosin head fragment linked via an oligopeptide linker to a target protein. Applicants' argument is not found persuasive.

While Kurzawa et al. do not specifically teach a linker between the myosin head fragment and the alpha-actinin repeats, clearly such linkers were well known in the art as evidenced by Bulow et al. and Argos. Moreover, the use of M765 was well characterized and established for use in the characterization of the structure, kinetics, and function of the myosin motor at the time of the invention. As such, one of ordinary skill in the art would have combined the teachings of the cited references to make the claimed invention.

[18] Claims 1, 3-4, 6, 8-10, and 13 are rejected under 35 U.S.C. 103(a) as being unpatentable over Manstein et al. (*J Mus Res Cell Mot* 16:325-332) in view of Bulow et al. and Argos. This rejection is necessitated by amendment. The claims are drawn to a recombinant protein comprising a first protein that is a motor domain of mysosin or an analog, fragment, or derivative thereof; a target protein of interest; and a linker between the first protein and target protein.

Manstein et al. teach a myosin fragment (amino acids 1-754 of *D. discoideum* myosin II) fused to one or two alpha-actinin repeats (see page 326, left column). Manstein et al. teach the fusion proteins have a histidine octamer at the C-terminus (page 326, left column). Manstein et al. do not specifically teach a linker between their fused protein domains.

At the time of the invention, the use of oligopeptide linkers for joining two protein components in a fusion protein to allow conformational flexibility of the protein components was well known to one of ordinary skill in the art. For example, Bulow et al. teaches the presence of peptide linkers between protein components of a fusion enzyme. Bulow et al. advises using linkers that are short (between two and ten amino acids) as being optimal as longer linkers are often prone to proteolytic cleavage and reduce recombinant protein yields (page 230, left column). Also, Argos teaches that oligopeptide linkers are used in the fusion of proteins and should generally not interfere with their function (page 943, left column). Argos teaches the advantages of using oligopeptide linkers comprising Ser, Gly, and Thr as these residues can impart flexibility and maintain stability and conformation in solution (page 956, left column).

Therefore, it would have been obvious to one of ordinary skill in the art to link the myosin head fragment (amino acids 1-754 of *D. discoideum* myosin II) and the alpha-actinin repeats of using a linker comprising at least two amino acids and glycine. One would have been motivated for the fusion protein of

Manstein et al. with a linker peptide comprising at least 2 amino acids and glycine because of the teachings of Bulow et al. and Argos. One would have a reasonable expectation of success for the fusion protein of Manstein et al. with a linker peptide comprising at least 2 amino acids and glycine because of the results of Manstein et al., Bulow et al., and Argos. Therefore, claims 1, 3-4, 6, 8-10, and 13, drawn to recombinant proteins as described above would have been obvious to one of ordinary skill in the art.

[19] Claim 12 is rejected under 35 U.S.C. 103(a) as being unpatentable over Manstein et al. (*J Mus Res Cell Mot* 16:325-332) in view of Bulow et al. and Argos as applied to claims 1, 3-4, 6, 8-10, and 13 above, and further in view of Manstein et al. (*Gene* 162:129-134). This rejection is necessitated by amendment. Claim 12 limits the linker of claim 1 to comprising Leu-Gly-Ser.

Manstein et al. (*J Mus Res Cell Mot* 16:325-332), Bulow et al., and Argos disclose the teachings as described above. Manstein et al. (*J Mus Res Cell Mot* 16:325-332) additionally teach that the plasmid used for expression of their fusion protein was derived from plasmid pDM-3H (page 326, left column). None of these references teaches a linker comprising the specific sequence of Leu-Gly-Ser.

Manstein et al. (*Gene* 162:129-134) teach an expression vector, pDXA-3H, with two restriction sites, KpnI and SacI, followed by nucleotides encoding Leu-Gly-Ser followed by XhoI and NsiI restriction sites.

Also, at the time of the invention, the use of inserting nucleic acids encoding a particular protein into an expression vector using a known restriction site present in the expression vector were well known in the art. The restriction sites KpnI, SacI, XhoI, and NsiI were well known restriction sites commonly used for insertion of nucleic acids into an expression vector at the time of the invention.

Therefore, it would have been obvious to one of ordinary skill in the art to combine the teachings of Manstein et al. (*J Mus Res Cell Mot* 16:325-332), Bulow et al., Argos, and Manstein et al. (*Gene* 162:129-134) for constructing an expression vector by inserting a nucleic acid encoding M754 into either the KpnI or SacI restriction sites of pDXA-3H and inserting a nucleic acid encoding the alpha actinin repeats into the XhoI and NsiI restriction sites of plasmid pDXA-3H to generate the fusion protein of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) with a linker comprising Leu-Gly-ser. One would have been motivated to construct an expression vector by inserting a nucleic acid encoding M754 into either

the KpnI or SacI restriction sites of pDXA-3H and inserting a nucleic acid encoding the alpha actinin repeats into the XhoI and NsiI restriction sites of pDXA-3H to generate the fusion protein of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) with a linker comprising Leu-Gly-ser in order to exploit the presence of restriction sites within vector pDXA-3H and to have a fusion protein with a linker comprising Gly and Ser as taught by Argos. One would have a reasonable expectation of success for constructing an expression vector by inserting a nucleic acid encoding M754 into either the KpnI or SacI restriction sites of pDXA-3H and inserting a nucleic acid encoding the alpha adinin repeats into the XhoI or NsiI restriction sites of pDXA-3H to generate the fusion protein of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) with a linker comprising Leu-Gly-ser because of the results of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) and Manstein et al. (*Gene* 162:129-134). Therefore, claim 12, drawn to the recombinant protein as described above, would have been obvious to one of ordinary skill in the art.

Applicants argues (beginning at page 11) that neither reference contains a suggestion regarding the desirability of making a recombinant protein comprising a myosin head fragment linked to a target protein that could be used to facilitate the purification and crystallization of the target protein of interest. Applicants' argument is not found persuasive.

Applicants intended use of the myosin head fragment, *i.e.*,to facilitate purification and crystallization of the target protein, is not a limitation in the claims. A recitation of the intended use of the claimed invention must result in a structural difference between the claimed invention and the prior art in order to patentably distinguish the claimed invention from the prior art. If the prior art structure is capable of performing the intended use, then it meets the claim. In this case, the limitation of "the purification and crystallization of which is desired" for the target protein in no way limits the target protein of part (b) of claim 1.

Applicants further argue the examiner has used hindsight reasoning to construct an obviousness rejection. Applicants' argument is not found persuasive.

In response to applicant's argument that the examiner's conclusion of obviousness is based upon improper hindsight reasoning, it must be recognized that any judgment on obviousness is in a sense necessarily a reconstruction based upon hindsight reasoning. But so long as it takes into account only

knowledge which was within the level of ordinary skill at the time the claimed invention was made, and does not include knowledge gleaned only from the applicant's disclosure, such a reconstruction is proper. See *In re McLaughlin*, 443 F.2d 1392, 170 USPQ 209 (CCPA 1971). In this case, the teachings disclosed by the prior art were within the level of ordinary skill at the time the claimed invention was made and do not include knowledge gleaned only from the applicant's disclosure. As such, the examiner properly combined the references.

[20] Claims 1, 3-4, 6, 8-13, and 30-31 are rejected under 35 U.S.C. 103(a) as being unpatentable over Manstein et al. (*J Mus Res Cell Mot* 16:325-332) in view of Ausubel et al. ("Current Protocols in Molecular Biology", John Wiley and Sons, Inc., 1995). This rejection is necessitated by amendment. The claims are drawn to recombinant proteins as described above.

Manstein et al. (*J Mus Res Cell Mot* 16:325-332) disclose the teachings as described above. In summary, Manstein et al. teach a fusion protein comprising *D. discoideum* myosin fused to alpha actinin with a C-terminal histidine octamer (page 331, right column). Manstein et al. further teach a rapid one-step purification of this fusion protein by depleting cellular ATP to form an insoluble complex with actin followed by elution with Mg2+-ATP (page 325, right column to page 326, left column and page 331, right column). Manstein et al. teach this method allows a rapid purification of recombinant myosins (abstract) and may also prove to be useful for the production of a wider range of proteins in *Dictyostelium,* particularly proteins that require post-translational modification (page 331, right column, top). Manstein et al. (*J Mus Res Cell Mot* 16:325-332) do not teach a linker comprising a factor Xa or thrombin protease cleavage site.

Ausubel et al. generally teach expression of fusion proteins (pages 16.4.1-16.4.3) Specifically, Ausubel et al. teach that it is advantageous to remove the carrier protein from the protein of interest to facilitate biochemical and functional analyses. Ausubel et al. teach enzymatic cleavage of the fusion protein is a desirable method and suggest factor Xa and thrombin as useful enzymes for such cleavage of a fusion protein (page 16.4.2, right column, bottom). Ausubel et al. teach the sequences of the factor Xa and thrombin cleavage sites (page 16.4.5).

Therefore, it would have been obvious to one of ordinary skill in the art to combine the teachings of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) and Ausubel et al. to modify the expression vector of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) to incorporate an encoding sequence for a factor Xa or thrombin protease cleavage site between the myosin and alpha-actinin components. One would have been motivated to incorporate a factor Xa or thrombin protease cleavage site between the myosin and alpha-actinin components of the fusion protein of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) in order to cleave the component proteins of the fusion following purification by the method of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) in order to remove the carrier protein from the protein of interest to facilitate biochemical and functional analyses as taught by Ausubel et al. One would have a reasonable expectation of success for incorporating a factor Xa or thrombin protease cleavage site between the myosin and alpha-actinin components of the fusion protein of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) because of the results of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) and Ausubel et al. Therefore, claims 1, 3-4, 6, 8-13, and 30-31, drawn to the recombinant protein as described above, would have been obvious to one of ordinary skill in the art.

[21] Claim 13 is rejected under 35 U.S.C. 103(a) as being unpatentable over Wolber et al. or Kuge et al. in view of Ford et al. (*Prot Exp Pur* 2:95-107). This rejection is necessitated by amendment. Claim 13 is drawn to the recombinant protein of claim 1 further comprising a tag sequence at the N- or C-terminus of the protein.

Wolber et al. disclose the teachings as described above. Wolber et al. further teach a method of purifying their fusion proteins by forming an insoluble precipitate in low salt and solvating the fusion protein in high salt followed by dialysis against low salt (page 900, right column to page 901, left column). Wolber et al. do not teach a tag at the N- or C-terminus of their fusion protein.

Kuge et al. disclose the teachings as described above. Kuge et al. further teach a method of purifying their fusion protein using column chromatography (page 1785, left column, top). Kuge et al. do not teach a tag at the N- or C-terminus of their fusion protein.

Ford et al. teach the use of fusion tails at the N- or C-terminus of a protein for increasing protein purity, promoting secretion, and providing useful assay tags (page 95, abstract).

Therefore, it would have been obvious to one of ordinary skill in the art to combine the teachings of Wolber et al. or Kuge et al. and Ford et al. to modify the expression vector of Wolber et al. or Kuge et al. to incorporate an encoding sequence for a fusion tag at the N- or C-terminus of the fusion protein of Wolber et al. or Kuge et al. One would have been motivated to incorporate a fusion tag at the N- or C-terminus of the fusion protein of Wolber et al. or Kuge et al. in order to increase the purity of the fusion protein, promote secretion of the fusion protein, or to provide an assay tag to rapidly identify those clones expressing the fusion protein. One would have a reasonable expectation of success for incorporating a fusion tag at the N- or C-terminus of the fusion protein of Wolber et al. or Kuge et al. because of the results of Wolber et al. or Kuge et al. and Ford et al. Therefore, claim 13, drawn to the recombinant protein as described above, would have been obvious to one of ordinary skill in the art.

[22] Claim 32 is rejected under 35 U.S.C. 103(a) as being unpatentable over Wolber et al. or Kuge et al. in view of Parks et al. (*Anal Biochem* 216:413-417). This rejection is necessitated by amendment. Claim 32 is drawn to the recombinant protein of claim 31 comprising a protease recognition sequence of ENLYFQG.

Wolber et al. and Kuge et al. disclose the teachings as described above. Neither Wolber et al. or Kuge et al. teach a protease recognition sequence of ENLYFQG in the linker of their respective fusion protein.

Parks et al. teach the use of the tobacco etch virus (TEV) nuclear inclusion a (NIa) protease for cleavage of fusion proteins. Parks et al. teach the TEV NIa cleavage site is ENLYFQG (page 414, Figure 1). Parks et al. teach TEV NIa cleavage of fusion proteins is an improvement over use of other proteases (abstract) because TEV NIa has many advantages including is its stability and activity over a range of pHs, ionic strengths, and temperatures, its specificity for its cleavage site, and lack of proteolysis of cryptic cleavage sites (page 416).

Therefore, it would have been obvious to one of ordinary skill in the art to combine the teachings of Wolber et al. or Kuge et al. and Parks et al. to modify the expression vector of Wolber et al. or Kuge et al. to replace the factor Xa cleavage site-encoding sequence with a TEV NIa protease cleavage site-encoding sequence between the myosin and target protein components. One would have been motivated

to incorporate a TEV NIa protease cleavage site between the myosin and target protein components of the fusion protein of Wolber et al. because TEV NIa cleavage of fusion proteins is an improvement over other proteases as taught by Parks et al. One would have a reasonable expectation of success for replacing the factor Xa cleavage site-encoding sequence with a TEV NIa protease cleavage site-encoding sequence between the myosin and target protein components because of the results of Wolber et al. and Parks et al. or to replace the thrombin cleavage site-encoding sequence with a TEV NIa protease cleavage site-encoding sequence between the myosin and target protein components because of the results of Kuge et al. and Parks et al. Therefore, claim 32, drawn to the recombinant protein as described above, would have been obvious to one of ordinary skill in the art.

[23] Claim 32 is rejected under 35 U.S.C. 103(a) as being unpatentable over Manstein et al. (*J Mus Res Cell Mot* 16:325-332) in view of Ausubel et al. ("Current Protocols in Molecular Biology", John Wiley and Sons, Inc., 1995) as applied to claims 1, 3-4, 6, 8-13, and 30-31 above and further in view of Parks et al. (*Anal Biochem* 216:413-417). This rejection is necessitated by amendment. Claim 32 is drawn to the recombinant protein of claim 31 comprising a protease recognition sequence of ENLYFQG.

Manstein et al. (*J Mus Res Cell Mot* 16:325-332) and Ausubel et al. disclose the teachings as described above. The references of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) and Ausubel et al. do not combine to teach a protease recognition sequence of ENLYFQG in a linker joining the components of the fusion protein of Manstein et al. (*J Mus Res Cell Mot* 16:325-332).

Parks et al. teach the use of the tobacco etch virus (TEV) nuclear inclusion a (NIa) protease for cleavage of fusion proteins. Parks et al. teach the TEV NIa cleavage site is ENLYFQG (page 414, Figure 1). Parks et al. teach TEV NIa cleavage of fusion proteins is an improvement over use of other proteases (abstract) because TEV NIa has many advantages including is its stability and activity over a range of pHs, ionic strengths, and temperatures, its specificity for its cleavage site, and lack of proteolysis of cryptic cleavage sites (page 416).

Therefore, it would have been obvious to one of ordinary skill in the art to combine the teachings of Manstein et al. (*J Mus Res Cell Mot* 16:325-332), Ausubel et al., and Parks et al. to modify the expression vector of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) to incorporate an encoding

sequence for a TEV NIa protease cleavage site between the myosin and alpha-actinin components. One would have been motivated to incorporate a TEV NIa protease cleavage site between the myosin and alpha-actinin components of the fusion protein of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) because TEV NIa cleavage of fusion proteins is an improvement over other proteases as taught by Parks et al. and in order to cleave the component proteins of the fusion following purification by the method of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) in order to remove the carrier protein from the protein of interest to facilitate biochemical and functional analyses as taught by Ausubel et al. One would have a reasonable expectation of success for incorporating a TEV NIa protease cleavage site between the myosin and alpha-actinin components of the fusion protein of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) because of the results of Manstein et al. (*J Mus Res Cell Mot* 16:325-332) and Parks et al. Therefore, claim 32, drawn to the recombinant protein as described above, would have been obvious to one of ordinary skill in the art.

Conclusion

[24] Status of the claims:

- Claims 1, 3-4, 6, 8-13, 28, and 30-32 are pending.
- Claims 1, 3-4, 6, 8-13, 28, and 30-32 are rejected.
- No claim is in condition for allowance.

THIS ACTION IS MADE FINAL. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to David Steadman, whose telephone number is (703) 308-3934. The Examiner can normally be reached Monday-Friday from 7:00 am to 5:00 pm. If attempts to reach the Examiner by telephone are unsuccessful, the Examiner's supervisor, Ponnathapura Achutamurthy, can be reached at (703) 308-3804. The FAX number for submission of official papers to Group 1600 is (703) 308-4242. Draft or informal FAX communications should be directed to (703) 746-5078. Any inquiry of a general nature or

relating to the status of this application or proceeding should be directed to the Art Unit receptionist whose telephone number is (703) 308-0196.

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